Untargeted metabolomics of human plasma with LC-EC-MS

Project background

Metabolism is a complex and dynamic process involving numerous metabolic pathways and enzymes, converting a wide range of nutrients, xenobiotics, and drugs, into energy, cellular building blocks, or waste. Quantitative analysis of metabolites is commonly performed by liquid chromatography (LC) coupled to mass spectrometry (MS). Many well-known metabolites are routinely analyzed, but there are numerous others which are missed by the standard targeted analysis methods. Untargeted metabolite analysis using high-resolution mass spectrometry is an emerging method in metabolomics, and allows differential analysis of 100s to 1000s of compounds, but identification of these compounds is not straightforward due to the limited compound-specific information obtained even with high resolution MS and MS/MS data.

Electrochemistry (EC) is an instrumental approach to oxidize redox-active compounds, including many metabolites and pharmaceuticals. In the Analytical Biochemistry group, we have previously developed and applied EC in combination with MS detection to generate and study drug metabolites, with the aim of understanding *in vivo* oxidative metabolism of drugs and xenobiotics by Cytochrome P450 enzymes in the liver. In addition, electrochemistry is well-known in the analytical field for the sensitive electrochemical detection (ECD) of electroactive species.

We are developing an on-line LC-EC-MS system to analyze and characterize metabolites and pharmaceutical drugs in biological samples, by combining EC and MS detection. Most importantly, EC oxidation adds a previously unexploited layer of data, yielding important information on both the redox activity (EC signal) and the oxidation products of each compound (MS signal) in the chromatogram. EC oxidation usually results in chemical transformation and thereby a mass change of a compound, e.g. by incorporation of an oxygen atom. The LC-EC-MS system has been applied as a proof of principle to fecal samples of patients with ADHD and treated with Ritalin [1]. Various differences related to drug intake we observed but more systematic analysis is needed.

Project tasks & goal

In this Master's project we will evaluate the merits of the LC-EC-MS method for analysis of (drug) metabolites in plasma samples. We will use plain plasma and plasma spiked with a variety of drug compounds to generate an untargeted metabolomics dataset on high resolution mass spectrometry instrumentation. Next, the sample will be analyzed with the electrochemistry cell coupled on-line with LC-MS and data sets at various cell potentials will be recorded. The combined data sets will be analyzed with available metabolomics software packages to determine the metabolite composition of the plasma samples. The spiked drug compounds and known metabolites in plasma (e.g. amino acids) will be used as references to track the overall performance of the method. Further optimization of EC parameters may be required to generate improved data sets.

[1] J Aresti Sanz. Electrochemistry meets mass spectrometry: a combined analytical platform for characterization of novel gut microbiota-produced metabolites in fecal samples. 2022. PhD Thesis. University of Groningen

The starting date is flexible. For more information contact:

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